

# A NEW SPECIES OF THE GENUS *RHEOBATRACHUS* (ANURA: LEPTODACTYLIDAE) FROM QUEENSLAND

by MICHAEL MAHONY\*, MICHAEL J. TYLER† & MARGARET DAVIES‡

## Summary

MAHONY, M., TYLER, M. J. & DAVIES, M. (1984) A new species of the genus *Rheobatrachus* (Anura: Leptodactylidae) from Queensland. *Trans. R. Soc. S. Aust.* **108**(3), 155–162, 13 December, 1984.

A leptodactylid frog *Rheobatrachus vitellinus* sp. nov. is described from the Clarke Range near Mackay, Queensland. The new species is larger than the gastric brooding frog *R. silus*, and is distinguished from it by a suite of external and internal characters. The two species are separated by a distance of approximately 800 km.

KEY WORDS. Anura, Leptodactylidae, chromosomes, *Rheobatrachus vitellinus* sp. nov.

## Introduction

The frog *Rheobatrachus silus* Liem (1973) was demonstrated by Corben *et al.* (1974) to be unique in the Animal Kingdom in brooding its young in its stomach, and eventually giving birth through the mouth. The histological and physiological modifications that accompany the conversion of a stomach to a brood sac have been the subject of extensive investigations based in Adelaide, and summarised by contributors to the volume edited by Tyler (1983). More recently other aspects of the cytology, pharmacology and physiology of gastric brooding have been documented by Gibbins & Tyler (1983), Tyler *et al.* (1983), Laidler *et al.* (1984), de la Lande *et al.* (1984) and Shearman *et al.* (1984).

In the light of the considerable interest in *R. silus*, the sudden demise of the population in 1979 assumed special significance. Despite extensive searches, particularly in 1982–83, not a single representative of *R. silus* has been found, but it is not possible to make a definite statement that the species is extinct.

In January 1984 a new species of *Rheobatrachus* was discovered in the Clarke Range in the vicinity of Eungella National Park, west of Mackay, Queensland, approximately 800 km north of the known geographic range of *R. silus*. Here we describe the new species.

## Materials and Methods

The specimens reported here are deposited in museum collections abbreviated as follows: AM Australian Museum, Sydney; QM Queensland Museum, Brisbane; SAM South Australian Museum, Adelaide.

Methods of measurement and abbreviations employed in the text follow Tyler (1968). Clearing and staining techniques for bone and cartilage are those of Dingerkus & Uhler (1977).

Mitotic chromosomes from one individual (sub-adult female QM J42145) were obtained from short term lymphocyte cultures using standard techniques applied to mammalian cultures (see Sharman *et al.* 1970), the only modification being incubation at 25°C. Whole blood (0.2–0.5 ml) was obtained by heart puncture using a sterile heparinized syringe.

## *Rheobatrachus vitellinus* sp. nov.

FIGS 1–11

*Holotype*: QM J42529, an adult female collected at Eungella National Park, 148°38'00"E.; 21°01'30"S., Queensland on 27.1.84 by K. R. McDonald and V. R. J. Hansen.

*Description of holotype*: Head flattened, approximately as long as broad. Snout not prominent, evenly rounded when viewed from above, rounded and projecting slightly in profile. Nares dorsal and inclined superiorly; surrounded by loose, fleshy margin and with small papilla at posterior border. Distance between naris and tip of snout approximately equal to eye to naris distance. Internarial span greater than distance separating eye from naris (E–N/IN 0.80). Canthus rostralis not defined. Eye prominent (Fig. 1).

Tympanum not visible externally. No vomerine teeth. Choanae large, widely separated and surrounded by rim of soft tissue. Tongue large and adherent to floor of mouth. Lower jaw with superior symphyseal prominence inserting into deep diastema between premaxillae. Upper jaw with medial innervation.

Fingers cylindrical with lateral fringe on medial surface of digit II. In order of length 3>4>2>1. Subarticular tubercles very poorly defined, no palmar tubercles. Slightly developed terminal discs

\* School of Biological Sciences, Macquarie University, North Ryde, N.S.W. 2113.

† Department of Zoology, University of Adelaide, Adelaide, S.A. 5001.

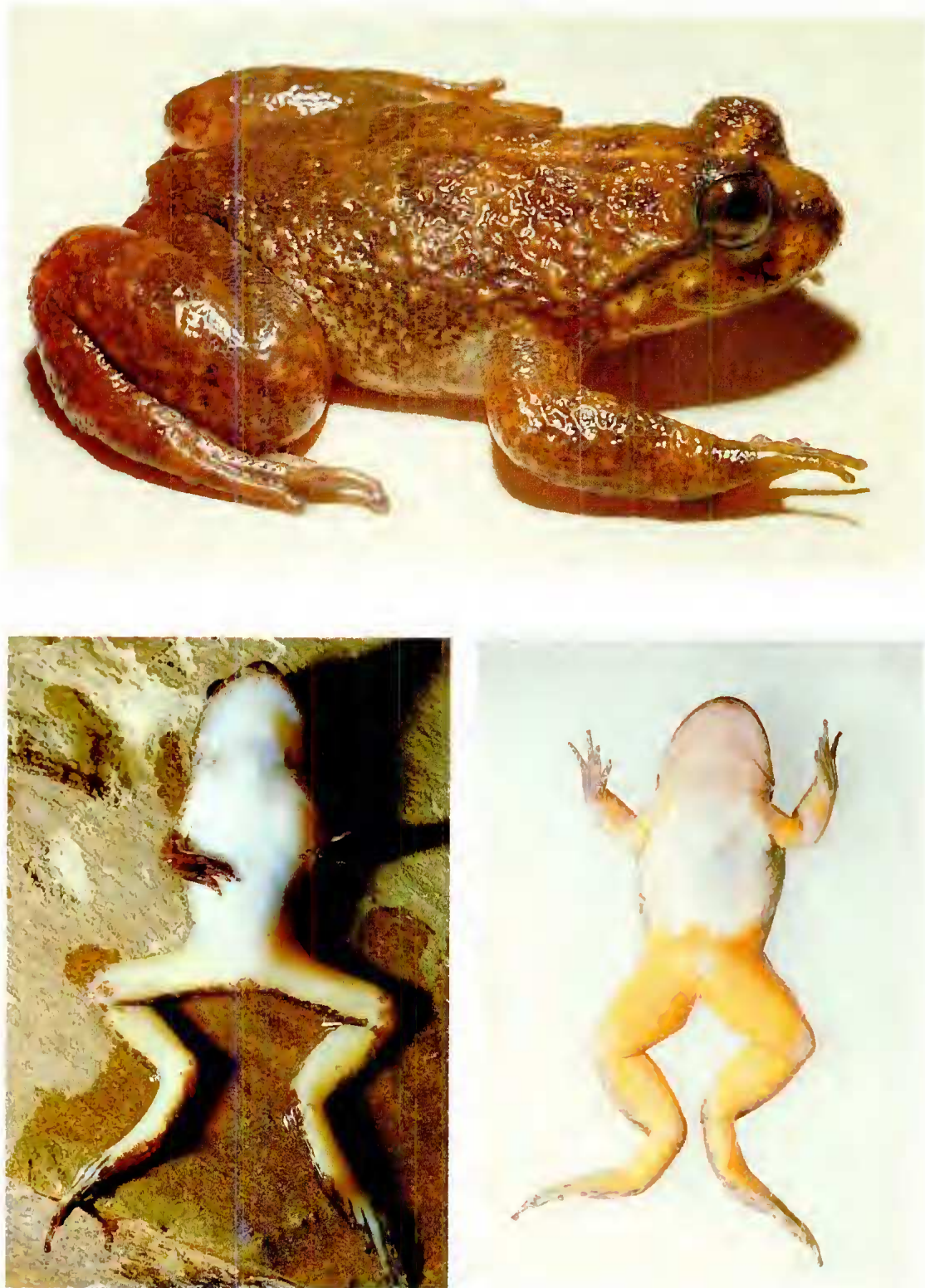


Fig. 1. Upper: Female *Rheobatrachus vitellinus* in life. Lower left: Ventral aspect of *R. silus*. Lower right: Ventral aspect of *R. vitellinus*.

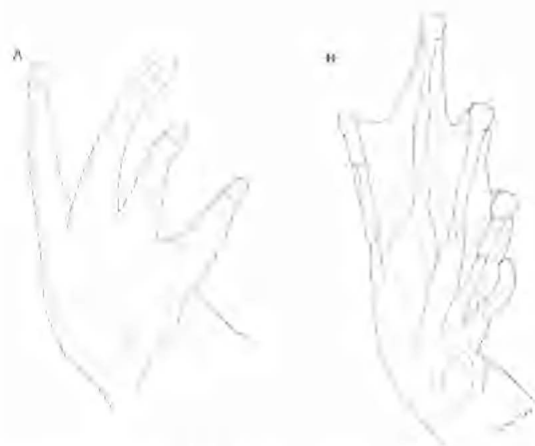


Fig. 2. *Rheobatrachus vitellinus*. A. Palmar surface of hand. B. Plantar surface of foot.

lacking circummarginal grooves. No interdigital webbing (Fig. 2A).

Hindlegs short (TL/S-V 0.47). Toes long, sub-articular tubercles moderately developed but lacking on metatarso-phalangeal joint of toe IV (Fig. 2B).

Expanded discs on tips of toes larger than those on fingers. Webbing reaching discs of all toes; broad medial flange on toe I. In order of length  $4 > 3 = 5 > 2 > 1$ . Large flattened inner but no outer metatarsal tubercle. No supernumerary tubercles on foot.

Dorsal skin irregular and foveolate. Large, irregular, bullate projections on upper eyelid. Distinct fold in customary supratympanic position extending from posterior corner of eye to insertion of forearm. Narrow, dermal fold superior to anterior  $\frac{1}{3}$  of supratympanic fold. No tarsal fold; few small, prominent tubercles on posterior surface of tarsus. No tubercles on flanks. Anus with fimbriated border. Ventral surface of body and limbs smooth.

After three months in preservative, dorsum mottled irregularly with dark brown and with small patches of cream upon a light brown background. Ventral surface cream with dense stippling of dark brown upon the throat and chest, but becoming more sparse posteriorly. Back of thighs heavily pigmented with dark brown. Palmar and plantar surfaces dark brown. Remainder of ventral surface pale cream.

#### Osteology:

##### Cranial Features

Skull poorly ossified; sphenethmoid poorly ossified, modified anterolaterally to form articulating surfaces, normally overlain by cartilaginous

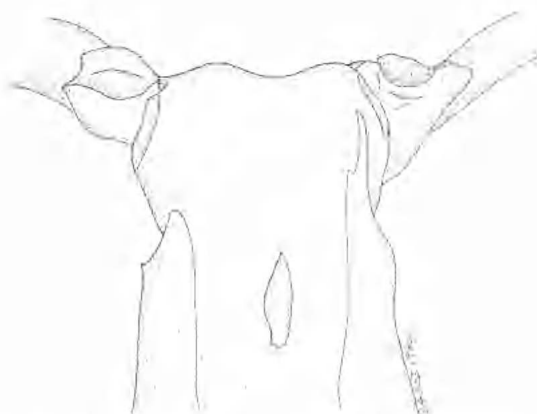


Fig. 3. Dorsal view of sphenethmoid of *Rheobatrachus vitellinus*.

cap (Fig. 3); not in bony contact with nasals, extending  $\frac{1}{4}$  length of orbit in ventral view. Elongate small bone located medially above sphenethmoid (Fig. 3). Prootic and exoccipital completely fused dorsally, slight reduction of ossification in plectral region. Crista parotica short and stocky, not articulating laterally with otic plate. Dorsally prootic extended posteromedially to form two flanges dorsolaterally to exoccipital. Frontoparietal fontanelle ovoid except for indented anterior extremity. Frontoparietals well ossified, anterior extremities slender, asymmetrical, not reaching anterior extremities of sphenethmoid. Orbital edges of frontoparietals straight, angled slightly posterolaterally. Anterior margins of frontoparietal fontanelle formed by sphenethmoid at level about  $\frac{1}{4}$  anteriorly along length of orbit. Posterior margin about  $\frac{3}{4}$  posteriorly along length of orbit (Fig. 4A).

Nasals small, slender, widely separated, expanded anteromedially, horizontal. Nasals not in bony contact with any roofing bones. Palatines broad, curved, unridged, overlying sphenethmoid ventrally to level of anterior extremities of frontoparietals (Fig. 4B). Parasphenoid moderately robust. Cultriform process short, tapering anteriorly, not reaching articulation of anterior ramus of pterygoid. Alary processes arising from ventral flanges in posteromedial region of cultriform process, short, moderately slender and crenate laterally.

Pterygoid extremely robust. Anterior rami in long contact with palatal shelf of maxillary. Medial rami extremely broad, blunt. Posteromedial flange at junction of three rami and ventromedial flange at posterior extremity of anterior ramus. Posterior rami moderately broad, long, acuminate. Quadratojugal robust and entire; squamosals robust with long acuminate zygomatic ramus and shorter expanded otic ramus.

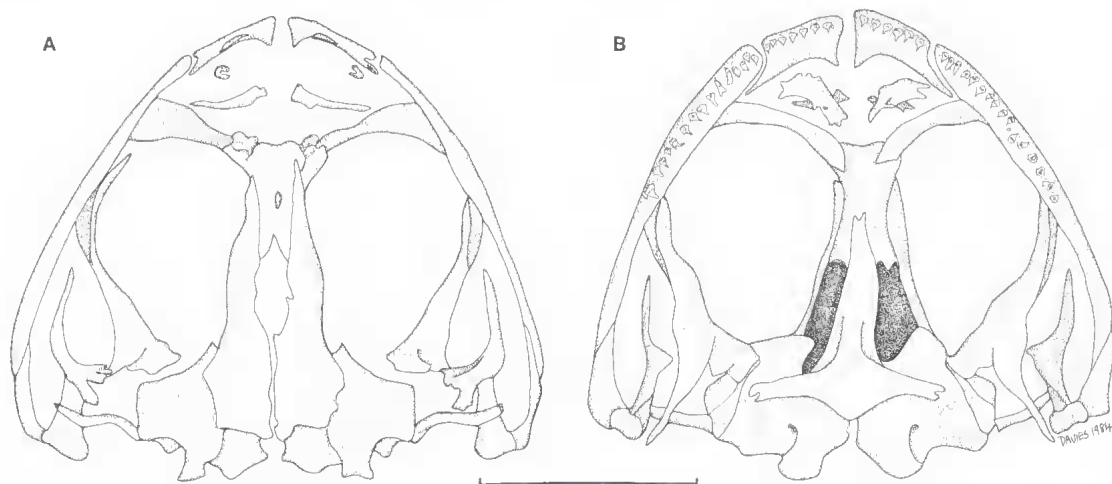


Fig. 4. Cranium of *Rheobatrachus vitellinus*. A. Dorsal aspect. B. Ventral aspect. Scale bar = 10 mm.

Maxillary and premaxillary dentate. Teeth fang-like. Pars facialis of maxillary shallow with well-developed preorbital process, not in bony contact with nasals. Alary processes of premaxillaries short, broad, perpendicular to pars dentalis of premaxillaries, but inclined medially. Palatal shelf well developed with poorly developed palatine processes of premaxillaries. Lateral extremities of palatal shelf of premaxillaries elongated to lie medially to anterior portion of palatal shelf of maxillaries (Fig. 4B). Pterygoid process not developed.

Vomers considerably reduced. Remnant denticulous processes present, but edentate. Alae poorly developed. Bony columella extremely long.

Ligaments joining mentomeckelian bones on lower jaw directed dorsally to form cartilaginous protuberance fitting into notch between palatine processes of premaxillaries. Meckel's cartilages poorly differentiated (Fig. 5C).

Hyoid plate broader than long. Alary processes with broad base, variable shape (Fig. 6). No aperture present on alary process for passage of hypoglossal nerve. Cricoid ring complete. Branchial processes simple, moderately long; oesophageal processes short. Posteromedial processes ossified; cartilaginous tips extended to lateral and medial extremities (Fig. 6).

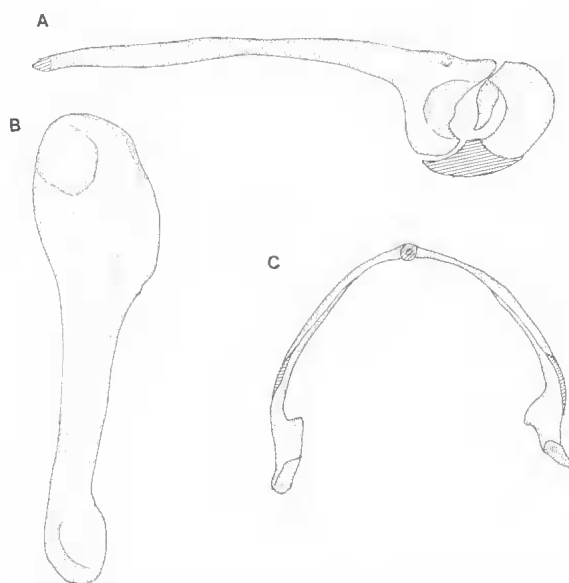


Fig. 5. *Rheobatrachus vitellinus*. A. Left ventral aspect of pelvic girdle. B. Humerus. C. Dorsal view of mandible.

#### Post Cranial Features

Pectoral girdle arciferal and robust (Fig. 7). Omosternum and ziphisternum present; xiphisternum  $\frac{1}{4}$  calcified. Clavicles robust, moderately separated medially. Coracoids robust, widely separated medially. Scapula bicapitate, very broad and stocky. Suprascapula about  $\frac{1}{4}$  ossified, proximal and posterolateral edges crenate (Fig. 7).

Well developed anteroproximal crest on humerus (Fig. 5B). Phalangeal formula of hand 2,2,3,3. Distal tips of phalanges knobbed. Six carpal elements present: radiale, ulnare, preaxiale centrale, postaxiale centrale, carpales 2 and 3. Bony prepollex (Fig. 8A).

Eight procoelous presacral vertebrae. Vertebra II imbricate, others non-imbricate (Fig. 9). Relative width of transverse processes III > IV > SD > II > V > VI > VII > VIII. Sacral diapophyses widely



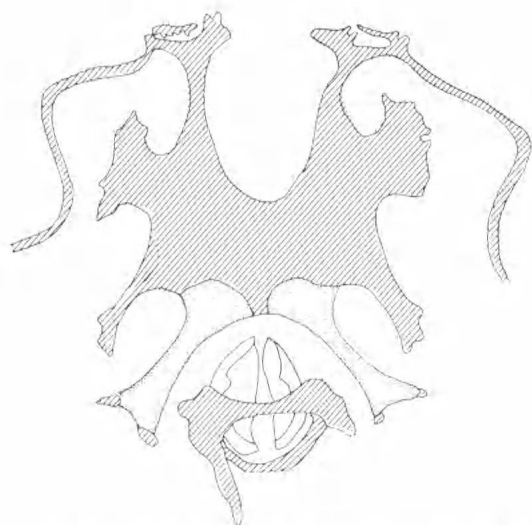


Fig. 6. Ventral view of hyoid plate and larynx of *Rheobatrachus vitellinus*.

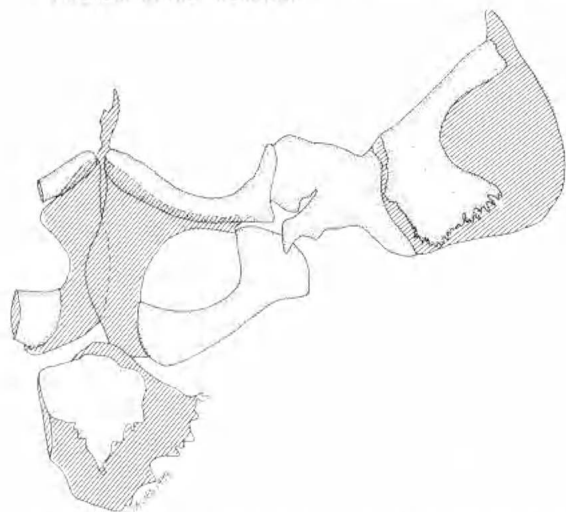


Fig. 7. Dorsal view of pectoral girdle of *Rheobatrachus vitellinus*.

expanded. Bicondylar sacrococcygeal articulation. (Fig. 9). Transverse processes present on urostyle. Urostyle long with poorly developed crest extending about  $\frac{1}{4}$  its length. Moderately developed dorsal prominence on ilium; tiny dorsal protuberance (Fig. 5A). Pubis cartilaginous.

Phalangeal formula of foot 2,2,3,4,3. Three distal tarsal elements present; O. fibulare and O. tibiare fused. Small bony prehallux (Fig. 8B).

No sesamoids present on appendages; free epiphysal joints not apparent.

**Variation:** There are four paratypes: AM R111733 an eviscerated adult male collected at Eungella National Park by K. R. McDonald and V. R. J.

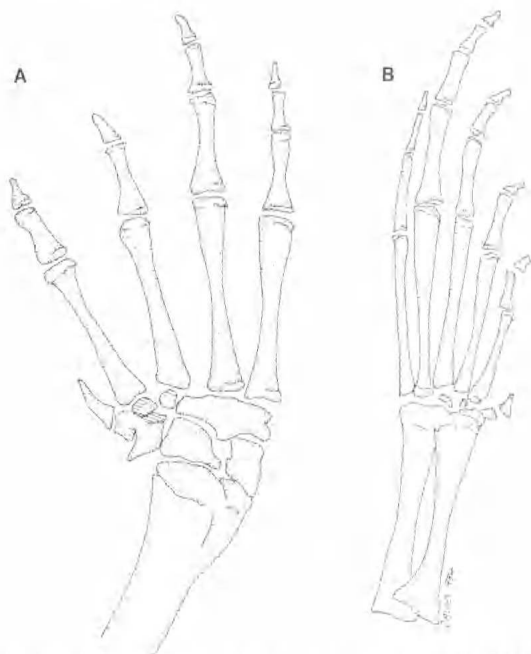


Fig. 8. *Rheobatrachus vitellinus*, bones of A. Hand. B. Foot.

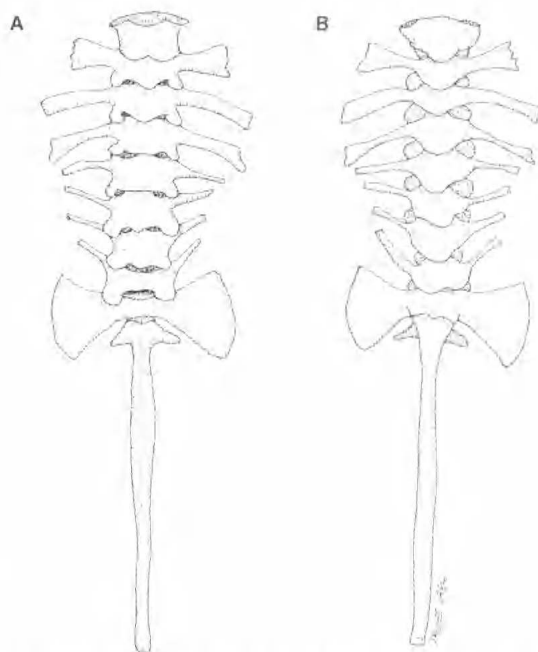


Fig. 9. *Rheobatrachus vitellinus*. Vertebral column. A. Dorsal aspect. B. Ventral aspect.

Hansen on 28.i.84; QM J42145 a sub-adult female collected at Tree Fern Creek, Clarke Range by M. Mahony on 2.i.84; SAM R25447 a cleared and stained adult female collected at Eungella National Park

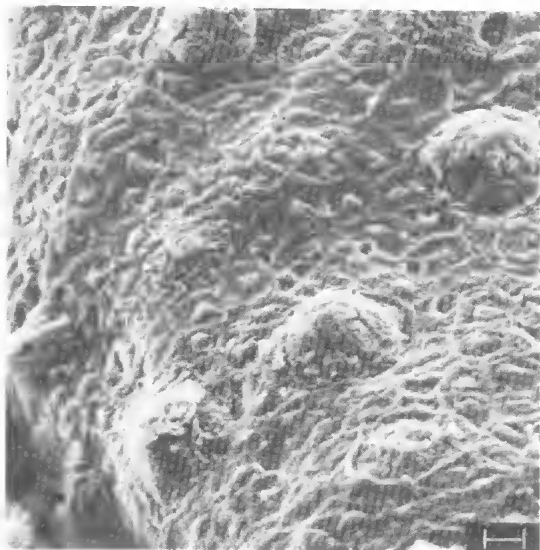


Fig. 10. *Rheobatrachus vitellinus*. Scanning electron micrograph of nuptial pad. Bar = 10  $\mu$  m.

by K. R. McDonald and G. Chester on 12.i.84; SAM R25446 an eviscerated adult female collected at Eungella National Park by K. R. McDonald and G. Chester on 10.i.84.

The adult females have S-V lengths of 68.9 mm and 62.2 mm respectively, the sub-adult female measures 41.5 mm and the adult male 55.7 mm.

In their habitus the paratypes do not differ significantly from the holotype. The head proportions are uniform; the eye is protuberant but the E-N/IN ratio is slightly higher (0.86–0.98) compared with 0.80 in the holotype.

The adult male has an unpigmented nuptial pad on the medial and dorsal surfaces of the first digit. With a magnification of 50x it is possible to see that

it is covered with numerous small spines (an SEM illustration is shown in Fig. 10). The male has a vocal sac with paired apertures on the floor of the mouth that are surrounded by fleshy margins.

**Colour in life:** All specimens examined by us have a pale brown dorsum with obscure darker patches on both the body and limbs (Fig. 1). The ventral surface of adults bears an extensive area of vivid yellowish-orange (Spectrum Orange of Smithe, 1975) covering the limbs and extending for varying distances up the abdomen. There also are patches of similar colour on the undersurface of the arms. The specimen illustrated (Fig. 1) has the remainder of the ventral surface unpigmented but in some individuals it is dark brown.

**Karyotype:** The karyotype of *R. vitellinus* is shown in Fig. 11; the diploid number is  $2n=24$ . The chromosomes are arranged in two groups on the basis of size. Pairs 1–6 are large with relative lengths (R.L.) ranging 15%–10%; pairs 7–12 are small with R.L. ranging 6%–3%. Chromosome pairs 1, 5, 7 and 8 are metacentric; pairs 3, 4 and 6 are submetacentric; pair 2 is subacrocentric; pairs 9–12 are acrocentric. A prominent secondary constriction occurs procentrically on the short arm of pair 6.

**Comparison with other species:** In appearance *R. vitellinus* differs from *R. silus* principally in its larger size and more spectacular ventral colouration. The three adult female paratypes of the new species have an S-V range of 62.2–68.9 mm (and the largest live specimen now in captivity is approximately 83 mm long). This size range compares with 44.5–53.9 mm S-V for 19 female *R. silus* examined by Tyler & Davies (1983). Similarly the sole adult male S-V of 55.7 mm compares with the cited range of 32.9–40.6 mm S-V for *R. silus*.

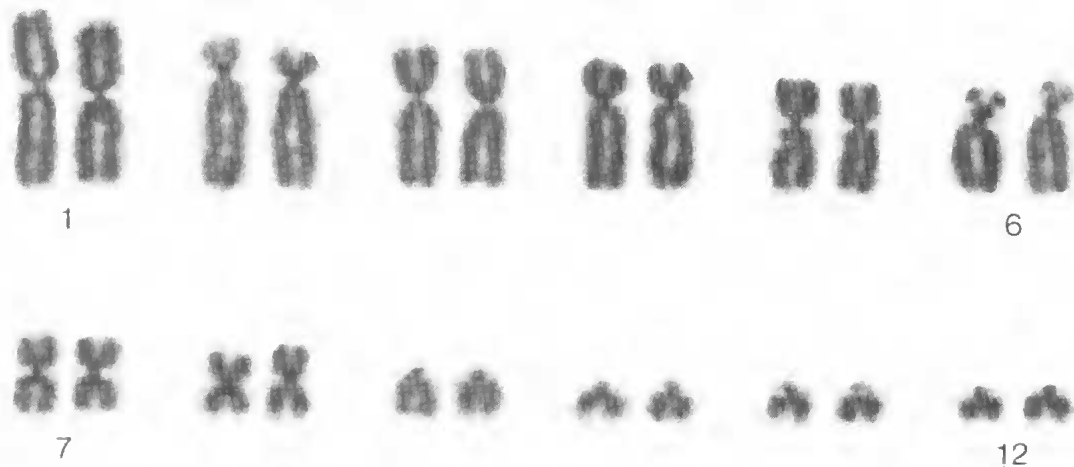


Fig. 11. Karyotype of *Rheobatrachus vitellinus*.

The striking difference in ventral colouration is shown in Fig. 1. *Rheobatrachus silus* has the ventral surface of the hindlimbs very pale yellow compared with the bright and more extensive yellow markings of *R. vitellinus*. No individual of *R. silus* exhibits the brown colouration seen in some, but not all, *R. vitellinus*.

Osteologically *R. vitellinus* differs from *R. silus* in a number of features. Cranially the presence of articulating facets anterolaterally on the sphenethmoid and of posteromedial flanges on the dorsal surface of the prootic, and the small supernumerary bone dorsal to the sphenethmoid, are unique to *R. vitellinus* and in fact are not shared by any other Australian leptodactylid. The position of the anterior extremities of the frontoparietals and posterolateral angle of the orbital edges of the frontoparietals differ between the two species. The skull of *R. vitellinus* is more extensively ossified in the crista parotica region and the frontoparietal fontanelle is less extensively exposed than in *R. silus* (Davies 1983).

Absence of the apertures for the hypoglossal nerve on the alary processes of the hyoid and minimal development of the mentomeckelian cartilages of the lower jaw are features unique to *R. vitellinus* in the genus.

Posteranally, the arciferal pectoral girdle of *R. vitellinus* (modified arciferal in *R. silus*) separates the two species, as does the crenate edges on the suprascapula and the relative widths of the transverse processes of the presacral vertebrae.

The karyotype of *R. vitellinus* is similar to those of the majority of Australian leptodactylid frogs, including *R. silus*, in diploid number and relative chromosome lengths. When compared with the karyotype of *R. silus* (Morescalchi & Ingram, 1974) differences are apparent in the centromere positions of several corresponding chromosome pairs and in the location of the secondary constriction. The most obvious differences in centromere position occur in pair 6 which is submetacentric in *R. vitellinus* and acrocentric in *R. silus*; pairs 9 and 10 are acrocentric in *R. vitellinus* and metacentric in *R. silus*. A prominent secondary constriction occurs procentrically on the short arm of pair 6 in

*R. vitellinus*. Morescalchi and Ingram (1974) did not identify any secondary constrictions in the karyotype of *R. silus*, however the acrocentric morphology of pair 6 means that a secondary constriction cannot possibly be in the same position as in *R. vitellinus*. These differences in chromosome morphology indicate that structural chromosomal rearrangements have occurred since the two species had a common ancestor and verify the specific identity of *R. vitellinus*.

**Habitat:** *Rheobatrachus vitellinus* is an aquatic species inhabiting shallow sections of fast flowing creeks in rain forest. Preliminary observations suggest that the species is confined to areas above approximately 300 m a.s.l. where the creeks flow across granitic rocks. K. R. McDonald currently is examining habitat preferences and distribution of the species.

In January 1984 the only other species of frogs observed at the creeks with *R. vitellinus* were *Toddactylus eungellensis* which was active on emergent rocks in the creek beds, and *T. liemi* which was calling from crevices in the creek bank.

**Etymology:** The specific name is derived from the Latin *vitellinus* 'of the yolk of an egg' and refers to the ventral colouration.

#### Acknowledgments

We record our gratitude to the Peter Rankin Trust Fund and the Australian Biological Resources Study for funding the visit by one of us (M.M.) which resulted in the discovery of this species. We are also deeply grateful to the Queensland National Parks and Wildlife Service for granting collecting permits, and particularly to Mr Keith McDonald for his enthusiastic cooperation, advice, and the provision of specimens. M.M. thanks S. Bergin, G. Johnston and P. Maidens for field assistance. A visit to the type locality by M.J.T. was made possible by the Australian Broadcasting Corporation. Laboratory studies were funded by a grant to M.J.T. and M.D. by the Australian Research Grants Scheme. The inclusion of colour plates was made possible by a grant from the Mark Mitchell Foundation.

#### References

- CORBEN, C. J., INGRAM, G. J. & TYLER, M. J. (1974) Gastric brooding: unique form of parental care in an Australian frog. *Science* **186**, 946-7.
- DAVIES, M. (1983) Skeleton. In M. J. Tyler (Ed.), *The Gastric Brooding Frog*, pp. 58-68. (Croom Helm: London and Canberra.)
- DE LA LANDE, I. S., O'BRIEN, P., SHEARMAN, D. J. C., TAYLOR, P. and TYLER, M. J. (1984) On the possible role of prostaglandin  $E_2$  in intestinal stasis in the gastric brooding frog *Rheobatrachus silus*. *Aust. J. Exp. Biol. Med. Sci.* **62**, 317-323.
- DINGERKUS, G. & UHLER, L. D. (1977) Enzyme clearing of alcian blue stained whole small vertebrates for demonstration of cartilage. *Stain Technol.* **52**, 229-231.

- GIBBINS, I. L. & TYLER, M. J. (1983) Changes in the organization and ultrastructure of smooth muscle cells in the stomach of the gastric brooding frog, *Rheobatrachus silus*, during brooding. *Cell Tissue Res.* **231**, 451-6.
- LAIDLER, P., TYLER, M. J. & SHEARMAN, D. J. C. (1984) The intestine of the gastric brooding frog *Rheobatrachus silus* during and after brooding: a morphological study. *Ibid.* (in press).
- LIEM, D. S. (1973) A new genus of frog of the family Leptodactylidae from SE Queensland, Australia. *Mem. Qld Mus.* **16**, 459-70.
- MORESCALCHI, A. & INGRAM, G. J. (1974) New chromosome numbers in Australian Leptodactylidae (Amphibia, Salientia). *Experientia* **30**, 1134-5.
- SHARMAN, G. B., ROBINSON, E. S., WALTON, S. M. & BERGER, P. J. (1970) Sex chromosomes and reproductive anatomy of some intersexual marsupials. *J. Reprod. Fertil.* **21**, 57-68.
- SHEARMAN, D. J. C., TAYLOR, P., TYLER, M. J., O'BRIEN, P., LAIDLER, P. & SEAMARK, R. F. (1984) An update on the role of prostaglandins in the stomach and intestine of the gastric brooding frog *Rheobatrachus silus*. In A. Allen, G. Flemström, A. Garner & W. Silen (Eds). "Mechanisms of mucosal protection in the upper gastrointestinal tract." (Raven Press: New York.)
- SMITHE, F. B. (1975) Naturalist's color guide. (American Museum of Natural History: New York.)
- TYLER, M. J. (1968) Papuan hylid frogs of the genus *Hyla*. *Zool. Verhand.* **96**, 1-203.
- (1983) (Ed.). "The Gastric Brooding Frog." (Croom Helm: London and Canberra.)
- , SHEARMAN, D. J. C., FRANCO, R., O'BRIEN, P., SEAMARK, R. F. & KELLY, R. (1983) Inhibition of gastric secretion in the gastric brooding frog, *Rheobatrachus silus*. *Science* **220**, 609-10.
- & DAVIES, M. (1983) Superficial features. In M. J. Tyler (Ed.). "The Gastric Brooding Frog", pp. 5-15. (Croom Helm: London and Canberra.)